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Short Communication

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Two- and Three-Dimensional Quantitative Structure-Activity Relationships Studies on a Series of Diuretics

Manish S. BHATIA [1]*, Kundan B. INGALE [1], Prafulla B. CHOUDHARI [1], Ramesh L. SAWANT [2] & Chandragouda R. PATIL [3]

[1] *Department of Pharmaceutical Chemistry, Bharati Vidyapeeth College of Pharmacy, Kolhapur, 416013, India*

[2] *Department of Pharmaceutical Chemistry, PDVVP'S College of pharmacy, Ahemadnager, India*

[3] *Department of Pharmacology, R.C. Patel College of Pharmacy, Shirpur, India*

***SUMMAARY*. Diuretics are an attractive class of drugs for the treatment of various disorders and in combination with some cardiovascular drugs. In the present work, 2D and 3D quantitative structure-activity relationship studies have been conducted on a series of diuretics. Significant correlation coefficients (r^2 = 0.81 and q^2 = 0.65, r^2 = 0.91 and q^2 = 0.85) were obtained, indicating potential of the models generated for untested compounds. The models were then used to predict the potency of an external test set, and the predicted values obtained from the 2D and 3D models were in good agreement with the experimental results. The final QSAR models, along with the information obtained from 3D steric and electrostatic contour maps and 2D contributions should be useful for the design of diuretics having improved potency.**

INTRODUCTION

A diuretic is a class of drug molecules which increases the excretion forms the kidneys; they are also used to clear fluid from the body in very critical conditions like heart failure where the body accumulates too much fluid. Diuretics are widely used in the treatment of diseases in wihich the fluids are accumulated in side the body. However after revealing their antihypertensive action many diuretics have acquired a new importance in medical practice and became basic agents for various forms and stages of hypertension, glaucoma, and other illnesses. Although the large research and development is going in the area of diuretic but not one of them is with out side effects, like ionic inbalance, hearing problem, and problem associated with diabetes, etc. The quantitative structure-activity relationship (QSAR) study provides medicinal chemists with the model to predict drug activity by mathematical equations which construct a relationship between the chemical structure and the biological activity. Now a days, a wide range of descriptors are being used in QSAR studies which can be classified into different categories including; constitutional, geometrical, topological, quantum, chemical and so on. There are different variable selection methods available including; multiple linear regression (MLR), genetic algorithm (GA), principal component or factor analysis (PCA/FA) and so on. The mathematical relationship between molecular descriptors and activity is generated to find the parameters affecting the biological activity and/or estimate the property of other molecules. The rapid increase in three-dimensional structural information (3D) of bioorganic molecules, coupled with the development of fast methods for 3D structure alignment, has led to the development of 3D structural descriptors and associated 3D QSAR methods. This information around the molecule is converted into numerical data using the partial least squares (PLS) method that reduces the dimensionality of data by generating components. Proceeding from this, the search for new potential diuretic agents with improved properties remains an urgent problem of pharmaceutical science. The main objective of our study was to investigate the quantitative structure-activity relationship (QSAR) of a series of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acid derivatives [1-5].

KEY WORDS: Diuretics, Drug design, QSAR.

*Author to whom correspondence should be addressed. *E-mail:* manish_bhatia13@yahoo.co.in

MATERIALS AND METHOD [6,7]

The structures of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic derivatives were synthesized as per reported literature and screened for their diuretic activity. The series of synthesized derivatives is shown in Table 1. The ED_{50} values were calculated and the data was usedf or QSAR studies..

QSAR Analysis (2D QSAR)

Data Set

The molecule builder module of Vlife MDS software was used to generate molecular models of series of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acid derivatives and further energy-minimized using the Merck Molecular Force Field (MMFF). Activity values for the QSAR equation were obtained using the negative logarithm of the effective concentration (EC). The physicochemical properties of each compound were specified using various descriptors. The selected descriptors include XAHydrophobicArea (XAHb), Most+ve&-vePotentialDistance (+,-PD), chiV3Cluster (cV3c), -vePotentialSurfaceArea (-PSA), QMDipoleX (QMDx), QMDipoleY (QMDy), QMDipoleZ (QMDz), SAHydrophilicArea (SAH), +vePotentialSurfaceArea (+PSA), SAHydrophobicArea (SAHb), YYPolarizability (YYP), Quadrupole3 (Q3), kappa3 (K3), highest occupied molecular orbital (HOMO), lowest unoccupied molecular orbital (LUMO), ionization potential (IP), heat of formation (Hf), radius of gyration (rgyr), molar refractivity (MR), molecular weight (Mwt.), partition coefficient (LogP). Twenty-one compounds from this data set were divided into training and test sets, the former set consisting of 16 randomly chosen compounds and the remaining in the latter set. The model developed using the training set was used to predict the activity of the compounds in the test set.

Full Search Multiple Linear Regression Method

Linear regression refers to the relationship between one or more variables denoted y and one or more variables denoted X, here in QSAR a relationship between independent and dependent variables physicochemical descriptors and biological activities respectively. Linear regression is achieved by fitting a best-fit straight line to the data using the least squares method. The F value represents the level of statistical significance of the regression. Quality of selected models was further ascertained to select the best

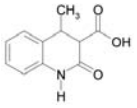
Sr. No	R	
1.	$PhCH_2$	
2.	$2\text{-}FC_6H_4CH_2$	
3.	$4\text{-}FC_6H_4CH_2$	
4.	$2\text{-}ClC_6H_4CH_2$	
5.	$4\text{-}ClC_6H_4CH_2$	
6.	$4\text{-}MeC_6H_4CH_2$	
7.	$2\text{-}MeOC_6H_4CH_2$	
8.	$4\text{-}MeOC_6H_4CH_2$	
9.	$3,4\text{-}(MeO)_2\ C_6H_3CH_2$	
10.	piperonyl	
11.	furfuryl	
12.	picolyl-2	
13.	picolyl-3	
14.	picolyl-4	
15.	PhCHMe	
16.	$PhCH_2CH_2$	
17.	$3\text{-}ClC_6H_4CH_2CH_2$	
18.	$4\text{-}ClC_6H_4CH_2CH_2$	
19.	$4\text{-}MeOC_6H_4CH_2CH_2$	
20.	$3,4\text{-}(MeO)_2C_6H_3CH_2CH_2$	
21.	$Ph(CH_2)_3$	

Table 1. Different substitutions performed in the 4-hydroxy-2-oxo-1,2- dihydroquinoline-3-carboxylic acid derivatives.

model from cross-validated squared correlation coefficient (q^2). The relation for q^2 is as shown below [**1**].

$$q^2 = 1 - \text{PRESS/TOTAL} \quad [\mathbf{1}]$$

$\sum(Y_{predicted} - Y_{observed})^2$ is the predictive error sum of squares (PRESS). $\sum(Y_{observed} - Y_{mean})^2$ is the total sum of squares (TOTAL), where $Y_{predicted}$, $Y_{observed}$, and Y_{mean} are the predicted, observed, and mean values of activity, respectively.

Given that, the full search method performs an exhaustive examination all possible descriptor combinations, there is little concern that important descriptors might be missed and this method enables identification of the QSAR equation with the best correlations. QSAR equations that have correlation coefficient which equal or exceed a preset value are reported. We specified 0.6 and 0.65 as the inter-correlation and correlation coefficient cutoff values, respectively. The selected models for various activities are shown in Table 2.

Activity prediction

To systematically assess a QSAR model, a reliable validation is required. Usually, a QSAR model is evaluated by the ability of the model to generate predicted results which have minimal deviation from the observed results of biological activity for the given dataset.

Ligand Preparation

The structure of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic was used as the template to built the molecules in the dataset in Vlife MDS 3.5. The ligand geometries were optimized by energy minimization using MMFF94 force field and Gasteiger-Marsili charges for the atoms, till a gradient of 0.001 kcal/mol/A° was reached, maintaining the template structure rigid during the minimization.

Molecular alignment

The molecules of the dataset were aligned by the atom-fit technique, using atoms common with the structure of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acid derivatives The most active molecule was selected as a template for alignment of the molecules. The alignment of all the molecules on the template is shown in Figure 1.

Descriptor Calculation

Like many 2D and 3D QSAR methods, a suitable alignment of given set of molecules was performed using the Vlife MDS 3.5 Engine. This was followed by generation of a common rectangular grid around the molecules. The hydrophillic, steric and electrostatic interaction energies are computed at the lattice points of the grid using a methyl probe of charge +1. These interaction energy values are considered for relationship generation and utilized as descriptors to decide nearness between molecules. The term descriptor is utilized in the following discussion to indicate field values at the lattice points.

Data Set

The dataset was divided into a training set (16 molecules, Table 1) and a test set (5 molecules, Table 1) on the basis of chemical and biological diversity using the random selection method for generation of the training and test set data. The effective concentration (EC) values for diuretic activity were used for the present QSAR study.

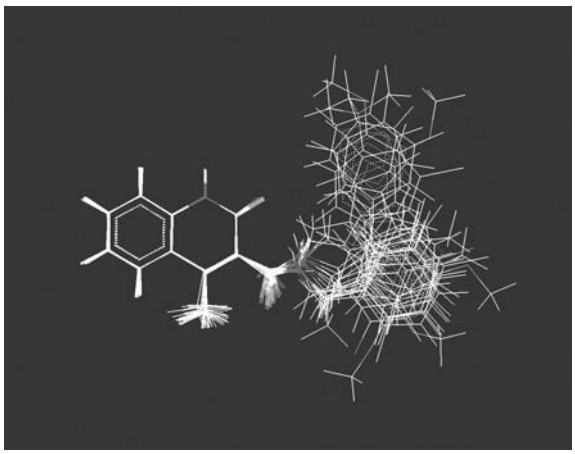


Figure 1. Alignment of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acid derivatives used in the QSAR model.

RESULTS AND DISCUSSION

In the present study, 21 molecules were used in the training set (Table 1) to derive QSAR models with the number of field grid points being not more than seven per model. To evaluate the predictive ability of generated 2D and 3D-QSAR models, a test set of 16 molecules with regularly distributed biological activities was used (Table 2). On successful runs of PLS, different sets of equations were generated by keeping the chain length of equations to seven, and these equations were further analyzed statistically to select the best model. As shown in Table 2, two models were selected after screening various combinations of different descriptors.

Interpretation of 2D QSAR Model

Model A best describes diuretic activity as confirmed by validation of the model judging internal and external predictivity and other statistical terms like the F value. The variable terms in the equation show low correlation among themselves, indicating a lesser probability of chance correlation. As indicated in Table 3, the activity is dependent on XAhydrophobic area, Heat of Formation and YYPolarizability XAhydrophobic area is vdW surface descriptor showing hydrophobic surface area calculated by Audry Method using Xlogp. This indicates the fraction of hydrophobic surface area of the total vdw surface area, which is available on the molecule. The total hydrophobic area is contribution of 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acid moiety as well as the substitutions carried out on the aromatic rings. The hydrophobic surface area is negatively correlated

Model	Equation	r^2	q^2	F value predicted	r^2 $r^2se^!$	PRED
Model A	pEC_{50} = 3.3591 -0.0061 (HeatOfFormation) +0.0054 (YYPolarizability) -4.4967 (XAAverage)	0.8112	0.6543	27.9260	0.5440	0.4028
Model B	pEC_{50} =4.6631+ 0.0232 E_571+6.4013 S_324-0.0534 E_901	0.9177	0.8548	72.4770	0.6112	0.4467

Table 2. Selected QSAR equations along with statistical parameters employed for model selection. ! Pred r^2 se is the mean of standard error in regression coefficient for the test set using the leave one out methodology.

Sr. No	2D QSAR(Model A)			3D QSAR(Model B)		
	observed	predicted	residuals	observed	predicted	residuals
1.	4.032915	4.066442	-0.033527	4.032915	4.179430	-0.146515
2.	4.237152	4.227910	-0.009242	4.237152	4.201754	0.035398
3.	4.309977	4.257829	-0.052148	4.309977	4.185029	0.124948
4.	4.372808	4.223621	-0.149187	4.372808	4.389061	-0.016253
5.	3.952263	4.057884	0.105621	3.952263	4.04970	-0.093757
6.	3.889641	3.889031	-0.000610	3.889641	3.916119	-0.026478
7.	3.900623	4.416883	0.516260	3.900623	4.025007	-0.124384
8.	4.369254	4.266986	-0.102268	4.369254	4.290449	0.078805
9.	4.187188	4.252646	0.065458	4.187188	4.145886	0.041302
10.	4.163249	4.248621	0.085372	4.163249	4.058376	0.104873
11.	4.362396	4.106299	-0.256097	4.362396	4.117680	0.244716
12.	4.042468	4.135074	0.092606	4.042468	4.526981	-0.484513
13.	4.156125	4.130082	-0.026043	4.156125	4.187911	-0.031786
14.	4.654436	4.28851	-0.485929	4.654436	4.671819	-0.017383
15.	3.944347	4.029519	0.085172	3.944347	3.920401	0.023946
16.	4.093102	4.113109	0.020007	4.093102	4.476591	-0.383489
17.	4.161443	4.141684	-0.019759	4.161443	4.293552	-0.132109
18.	4.374903	4.244094	-0.130809	4.374903	4.300079	0.074824
19.	4.062625	4.083403	0.020778	4.062625	4.056666	0.005959
20.	4.776869	4.813235	0.036366	4.776869	4.772005	0.004864
21.	4.416659	4.068320	-0.348339	4.416659	4.326957	0.089702

Table 3. Observed and predicted activity by QSAR equations along with the residuals.

with the activity. This indicates that the molecules possessing greater hydrophobic surface area will have reduced activity. The reason for this may be that increase in hydrophobic surface area may lead to increase in size of the molecule, which may leads to transport through various channels or pores to the target. Reduction in hydrophobic surface can be achieved by adding hydrophilic or polar substituents along the .quinoline nucleus. YY polarizablity Induced polarizability along YY axis. The receptor interaction required for the diuretics activity favour the polarizablity of synthesized molecules. Thus the parameter is positively correlated to the biological activity. Heat of formation is a semi- empirical type of descriptor showing negative correlation with the biological activity.

Interpretation of 3D QSAR Model

The model B describes the structural features optimum for the diuretic activity. The steric and electrostatic fields were calculated using the MMFF94 force field and Gasteiger-Marsili charges. A training set of 16 molecules, and a test set of 5 molecules was used as described earlier. The model was selected on basis of r^2, q^2, pred r^2, F and p values. The r^2 value for model A was 0.9235. The F test and p significance values were considered for the selection of model.

The points that were found optimum for the activity after the QSAR study are shown in Figure 2. The contribution of points E_571, S_324 which are the electronic and steric interaction fields at lattice points 571, 324 along with points E_901 which are the electrostatic interaction field at lat-

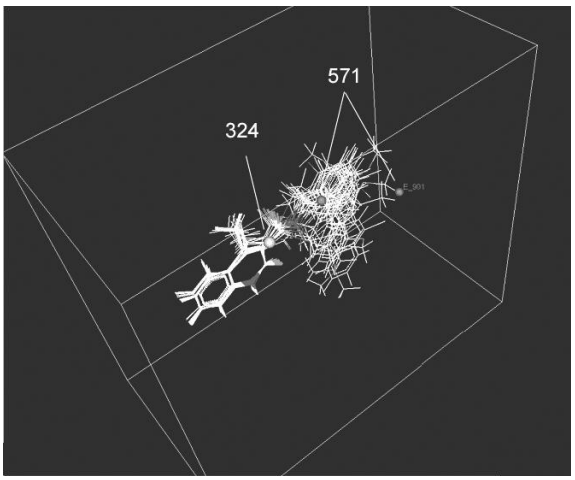


Figure 2. Field points used in the QSAR model.

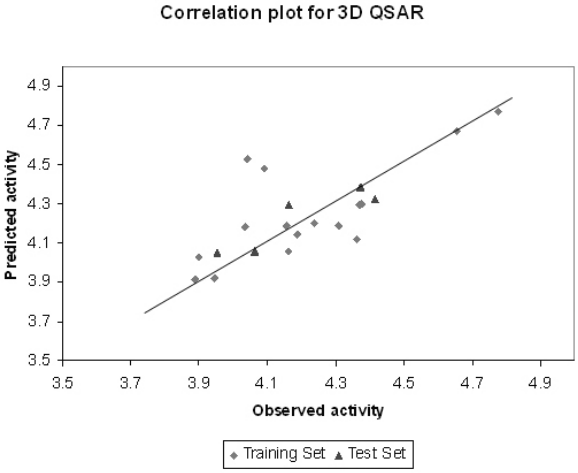


Figure 4. Correlation plot for 3D QSAR.

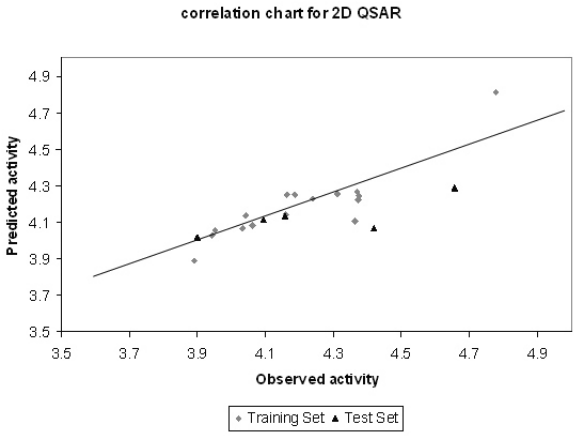


Figure 3. Correlation plot for 2D QSAR.

tice points 901, imply that these points are indeed significant for the structure-activity relationship. The positive contribution of the fields S324, E571 indicates that the addition of groups having steric interaction at lattice point 324 and groups having electrostatic interactions at lattice points 571 are required for amplified diuretic activity. Along with this the fields E 901which contribute negatively to the activity also need to be taken into account and need to be reduced.

CONCLUSIONS

The 3D and 2D QSAR statistical models described in this work (Figs. 3 and 4) show both good internal and external consistency, and represent important contribution to the QSAR field in the area of designing novel diuretic drugs. The good correlation between experimental and predicted pEC50 values for the test set compounds further proved the reliability of the constructed QSAR models. It is worth noting that we have employed the same training and test sets for all QSAR analyses, and the results showed that investigations can be carried out. From the 3D QSAR studies steric and electrostatic contour maps we can conclude that electronegative groups surrounding the 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acid moiety are related to improved potency. In addition, the favorable steric contours suggest that aromatic bulky groups at the 4-hydroxy-2-oxo-1,2-dihydroquinoline-3-carboxylic acid moiety may increase ligand potency. The 3D and 2D QSAR models should be useful for the design of new structurally related potential diuretics having improved affinity and potency. Thus the integration of 3D and 2D QSAR methods is will act as a important tool in medicinal chemistry and drug design studies.

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